

520 Rec'd PCT/PTO 29 OCT 1999

FORM PTO 1390 (REV. 5-93)	U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE	ATTORNEY DOCKET NUMBER 2534-00053
TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371		U.S. APPLICATION NO. (if known, 37 C.F.R. 1.1) 09/423004

INTERNATIONAL APPLICATION NO. PCT/FI98/00370	INTERNATIONAL FILING DATE 28 April 1998	PRIORITY DATE CLAIMED 29 April 1997
---	--	--

TITLE OF INVENTION
METHOD FOR THE MATURATION OF BEER

APPLICANT(S) FOR DO/EO/US
Matti LINKO; Ilkka VIRKAJARVI; Jukka KRONLOF; Esko PAJUNEN

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

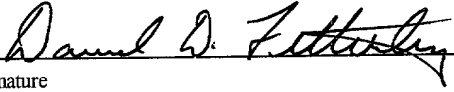
1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371.
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☐ This express request to begin national examination procedures (35 U.S.C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and PCT Articles 22 and 39(l).
4. ☐ A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.
5. ☐ A copy of the International Application as filed (35 U.S.C. 371(c)(2))
 - a. ☐ is transmitted herewith (required only if not transmitted by the International Bureau).
 - b. ☐ has been transmitted by the International Bureau.
 - c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☐ A translation of the International Application into English (35 U.S.C. 371(c)(2)).
7. ☐ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))
 - a. ☐ are transmitted herewith (required only if not transmitted by the International Bureau).
 - b. ☐ have been transmitted by the International Bureau.
 - c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
 - d. ☐ have not been made and will not be made.
8. ☐ A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. ☐ An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. ☐ A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11. to 16. below concern other document(s) or information included:

11. ☐ An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
12. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. ☒ A FIRST preliminary amendment.
☐ A SECOND or SUBSEQUENT preliminary amendment.
14. ☐ A substitute specification.
15. ☐ A change of power of attorney and/or address letter.
16. ☒ Other items or information:

Supplement to Transmittal Letter

[illegible]

U.S. APPLICATION NO. (If known, see P. C. 1.5) 09/423004	INTERNATIONAL APPLICATION NO. PCT/FI98/00370	ATTORNEY'S DOCKET NUMBER 2534-00053
CERTIFICATE OF EXPRESS MAIL		
<p>I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as EXPRESS MAIL-POST OFFICE TO ADDRESSEE, in an envelope addressed to: BOX PCT, COMMISSIONER OF PATENTS AND TRADEMARKS, WASHINGTON, D.C. 20231 on the <u>29th</u> day of October, 1999. Express Mail Label <u>EL269044505US</u>.</p>		
Daniel D. Fetterley	20,323	
Name	Reg. No.	
	10/29/99	
Signature	Date	

09/423004

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Application of)	Group Art Unit:
)	
MATTI LINKO ET AL)	Examiner:
)	
Int'l. Appln. No. PCT/FI98/00370)	METHOD FOR THE MATURATION
)	OF BEER
Int'l. Filing Date: 28 Apr 1998)	

PRELIMINARY AMENDMENT

Milwaukee, Wisconsin 53202
October 29, 1999

Box PCT Application
Asst. Commissioner for Patents
Washington, D.C. 20231

Sir:

Prior to computing the filing fee in this application, kindly amend the above identified application, as follows. The filing fee is to be computed on the amended claims.

IN THE ABSTRACT:

Cancel the Abstract presently in the application and substitute therefor the Abstract attached to this Preliminary Amendment.

IN THE SPECIFICATION:

Page 1, after the title and before the first line of text, insert the heading ---
BACKGROUND OF THE INVENTION---

Page 4, between lines 14 and 15, insert the heading ---SUMMARY OF THE
INVENTION---

Page 4, between lines 30 and 31, insert the heading ---DETAILED
DESCRIPTION OF THE INVENTION---

Page 4, line 33, delete "claim 1" and substitute therefor ---the claims---

Page 4, line 35, delete "claim 13" and substitute therefor ---the claims---

Page 6, line 13, delete "specification"

IN THE CLAIMS:

Claim 3, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1---

Claim 3, line 2, delete "2"

Claim 4, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1---

Claim 4, line 2, delete "2"

Claim 5, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1---

Claim 5, line 2, delete "2"

Claim 6, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1---

Claim 6, line 2, delete "5"

Claim 7, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1"

Claim 7, line 2, delete "6"

Claim 8, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1---

Claim 8, line 2, delete "7"

Claim 9, line 1, delete "any one of claims 1 -" and substitute therefor ---claim

1---

Claim 9, line 2, delete "8"

Claim 10, line 1, delete "any one of claims 1" and substitute therefor ---claim

1---

Claim 10, line 2, delete "- 9"

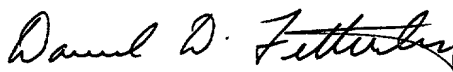
Claim 11, line 1, delete "any one of claims 1" and substitute therefor ---claim

1---

Claim 11, line 2, delete "- 10"

Respectfully submitted,

ANDRUS, SCEALES, STARKE & SAWALL, LLP



Daniel D. Fetterley
(Reg. No. 20,323)

100 East Wisconsin Avenue, Suite 1100
Milwaukee, Wisconsin 53202
(414) 271-7590
Atty. Docket No. 2534-00053 (C.11369-120)

CERTIFICATE OF EXPRESS MAIL

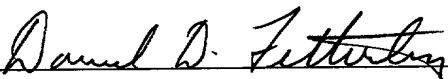
I hereby certify that this correspondence is being deposited with the United States Postal Service, with sufficient postage, as EXPRESS MAIL - POST OFFICE ADDRESSEE, in an envelope addressed to: Box PCT Application, Asst. Commissioner for Patents, Washington, D.C. 20231 on the 29th day of October, 1999. The Express Label is EL269044505US.

Daniel D. Fetterley

20,323

Name

Reg. No.



Signature

10/29/99

Date

ABSTRACT OF THE DISCLOSURE

The invention relates to a continuous method for the maturation of beer after main fermentation, in which method the unmatured beer, after the removal of yeast and a heat treatment, is passed into a bio-reactor filled with a carrier material with yeast
5 immobilized on it, said carrier material mainly consisting of wooden particles and/or similar particles. The invention also relates to a continuous maturation reactor, which is an upright column-type flow-through reactor containing one or more sieves, intermediate bottoms or flanges and which is filled with a carrier material with yeast immobilized on it, said carrier material mainly consisting of wooden particles and/or similar particles.

09 / 423 004

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Application of)	Group Art Unit:
)	
MATTI LINKO ET AL)	Examiner:
)	
Int'l. Appln. No. PCT/FI98/00370)	METHOD FOR THE MATURATION OF
)	BEER
Int'l. Filing Date: 28 Apr 1998)	

SUPPLEMENT TO TRANSMITTAL LETTER


Box PCT Application
Asst. Commissioner for Patents
Washington, D.C. 20231

Sir:

Entry of this application into the National Phase in the United States is pursuant to 37 C.F.R. §1.494(c)/1.495(c). The papers include an English language specification and claims, and the drawing. The oath or declaration of the applicant is not included. The declaration will be forwarded promptly upon notification from the U.S. Patent and Trademark Office.

Respectfully submitted,

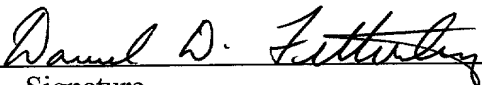
ANDRUS, SCEALES, STARKE & SAWALL, LLP


Daniel D. Fetterley
(Reg. No. 20,323)

100 East Wisconsin Avenue, Suite 1100
Milwaukee, Wisconsin 53202
(414) 271-7590
Atty. Docket No.: 2534-00053 (C.11369-120)

CERTIFICATE OF EXPRESS MAIL

I hereby certify that this correspondence is being deposited with the United States Postal Service, with sufficient postage, as EXPRESS MAIL - POST OFFICE TO ADDRESSEE, in an envelope addressed to: Box PCT Application, Asst. Commissioner for Patents, Washington, D.C. 20231 on the 29th day of October, 1999. The Express Mail label number is EL269044505US.

<u>Daniel D. Fetterley</u>	<u>20,323</u>
Name	Reg. No.
	<u>10/29/99</u>
Signature	Date

WO 98/49264

09/423004
PCT/FI98/00370

1

420 Rec'd PCT/PTO 29 OCT 1999

METHOD FOR THE MATURATION OF BEER

The present invention relates to a continuous method for the maturation of beer after main fermentation, in which method the unmaturred beer, after removal of yeast and a heat treatment, is passed into a bio-reactor filled with a carrier with yeast immobilised on it. The invention also relates to a continuous maturation reactor, which is an upright column-type flow-through reactor containing one or more
10 sieves, intermediate floors or flanges and which is filled with a carrier with yeast immobilised on it.

Beer production generally comprises the following main steps:

- 15 malting of grain (usually barley) by germinating,
 crushing of the malted grain to produce malt grist,
 adding water into the grist to form a mash,
 mashing to decompose starch into fermentable
20 sugar,
 separating the wort thus produced from the mash,
 cooking the wort with hops to produce a taste and aroma and to stop the enzymatic activity,
25 clarifying and cooling the wort,
 fermenting the wort with yeast to convert the glucose and maltose into ethanol and carbon dioxide (main fermentation) to produce unmaturred beer,
 maturing the unmaturred beer (secondary fermentation), and
30 filtering and stabilising the beer and putting it into suitable containers.
- 35 The maturation of beer is an important operation to give the beer a mellow and homogeneous taste and flavour.

00423004 042400

WO 98/49264

PCT/FI98/00370

2

Traditionally, beer is matured by storing the un-
matured beer for several weeks at a low temperature
after the main fermentation. This involves high stor-
age costs, which has given rise to the development of
5 a fast continuous method for the maturation of beer to
substitute storage. In this method, the yeast is re-
moved from the unmatured beer after the conventional
main fermentation, the unmatured beer is subjected to
a heat treatment (e.g. 80 - 90 °C for 5 - 15 min),
10 whereupon the beer is cooled (e.g. 10 - 15 °C) and
then matured in a reactor in which the yeast is immo-
bilised on a carrier. Finally, the beer is finished,
i.e. stabilised and filtered in the conventional man-
ner. The retention time in the continuous reactor is
15 of the order of e.g. two hours.

During the heat treatment, the α -acetolactate
contained in the unmatured beer is converted to diace-
tyl and partly also acetoin. The taste of diacetyl is
felt in beer even when the acetyl concentration is
20 only 0.05 mg/l. It is a strong sugary or taffy-like
taste and flavour, which is characteristic of unma-
tured or newly brewed beer. In the reactor, the yeast
reduces the diacetyl into acetoin. At the same time,
certain other carbonyl compounds are also reduced, and
25 the result is a savoury beer. Acetoin has a milder
taste and flavour, and the threshold concentration, 50
- 1000 mg/l, above which its taste is felt in beer is
considerably higher than for diacetyl.

Prior-art methods are described e.g. in the
30 following articles: Monograph XXIV of the European
Brewery Convention, E.B.C.-Symposium Immobilized yeast
applications in the brewing industry, Espoo, Finland,
October 1995 (ISBN 3-418-00749-X); E. Pajunen: Immo-
bilized yeast lager beer maturation: DEAE-cellulose at
35 Sinebrychoff (pages 24-40) and I. Hyttinen: Use of po-
rous glass at Hartwall brewery in the maturation of
beer with immobilized yeast (pages 55-56). In the for-

00443004-0400

WO 98/49264

PCT/FI98/00370

3

mer application, the carrier used to immobilise the yeast is DEAE cellulose with titanium dioxide and polystyrene mixed in it; patent specification US 4915959 describes the same application. In the latter
5 application, the carrier is porous glass. In the production of beer containing only a small amount of alcohol or no alcohol, a column in which yeast is immobilised in DEAE cellulose (H.Lommi: Immobilized yeast for maturation and alcohol-free beer, Brewing and Dis-
10 tilling International, May 1990, pp. 22-23) has been used.

These applications work well in a technical sense, and the beer produced is of good quality, the same as beer matured by the traditional method. How-
15 ever, the problem with the known applications is the high cost of the carrier materials. Purchase of the carrier material is a significant investment, and because of the high price the carrier must be regenerated after a certain period of use so that it can be
20 used again.

In traditional maturation in a container, fairly large wooden strips e.g. 400 - 500 mm long and 40 - 50 mm wide have been added into the storage con-
25 tainers. The purpose of the strips is to bind some of the yeast and thus to promote the clarification, and to some extent, secondary fermentation of the beer. This is a conventional slow batch process. Some breweries still use this procedure, mainly to preserve the tradition.

In the production of ethanol by a continuous fermenting process, immobilisation of yeast has been effected by using pieces of wood, e.g. beech, (M. Moo-
30 Young, J. Lamptey and C.W. Robinson: Immobilisation of yeast cells on various supports for ethanol production, Biotechnology Letters 2 (1980) No. 12, pp. 541-
35 545) and birch (M.A. Gencer and R. Mutharasan: Ethanol fermentation in a yeast immobilised tubular fermentor,

0044004 0440

WO 98/49264

PCT/FI98/00370

4

Biotechnology and Bioengineering 25 (1983) 2243-2262).
However, the production of ethanol is completely different from the manufacture of beer: in the former, the aim is to achieve a fermenting process as effective as possible, whereas in the latter the primary objective is to develop the desired good taste and flavour in conjunction with the fermenting process.

In the production of beer, small-scale experiments have also been carried out in which wooden chips have been used in conjunction with main fermentation to immobilise yeast: J. Kronlöf and V.-P. Määttä: Main fermentation using immobilised yeast in beer production, Mallas ja Olut 1993, No. 5, pp. 133-147).

The object of the present invention is to eliminate the drawbacks mentioned above.

The object of the invention is to disclose a fast, continuous method for the maturation of beer, in which yeast immobilised on a carrier effectively reduces the diacetyl concentration to a level below an acceptable taste threshold and which is applicable for use in conjunction with known beer production methods for the maturation of unmaturred beer.

Another object of the invention is to disclose a fast, continuous method for the maturation of beer in which the carrier is an economically priced and risk-free material.

A further object of the invention is to disclose a continuous maturation reactor for implementing the method.

The method of the invention for the maturation of beer is characterised by what is presented in claim 1.

The maturation reactor of the invention is characterised by what is presented in claim 13.

The invention is based on research work carried out, the aim of which was to apply the technique

04123004 043400

WO 98/49264

PCT/FI98/00370

5

of immobilising yeast to secondary fermentation and maturation of beer. It was unexpectedly established that wooden particles and/or similar particles are excellently suited for use as a carrier for the immobilisation of yeast.

In the continuous method of the invention for the maturation of beer, the unmaturred beer, after the removal of yeast and a heat treatment, is passed into a bio-reactor filled mainly with wooden particles and/or similar particles with yeast immobilised on them. The principle of the method of the invention is the same as in industrial procedures using DEAE cellulose or porous glass as a carrier. The yeast removal and other secondary treatment operations are performed as in the known procedures.

The method of the invention is applicable for the production of various kinds of beer, i.e. bottom yeast beer and scum yeast beer. Suitable raw materials are malt and other sources of starch and sugar as are known in beer production. The beer to be produced may have an alcoholic content between 0 - 10 % and a pitching wort content between 5 - 20 % or more, even 30 %.

In the method of the invention, the carrier may consist of wooden particles and/or similar particles of any size and shape, preferably cut into fairly small chips, sticks or into the shape of any regular or irregular bodies of roughly uniform size. The largest dimension of the particles is mainly 1 - 100 mm, advantageously 1 - 50 mm and preferably 2 - 20 mm.

The wooden particles to be used may be produced from any deciduous wood species, e.g. aspen, beech, palm or the like. The particles may also be produced from coniferous wood. The wood species to be used can be so chosen that the aromatic substances contained in it will have a desired effect on the taste and flavour of the beer to be produced. The par-

09423004 042400

WO 98/49264

PCT/FI98/00370

6

ticles may also be produced from tropical grass, e.g. bamboo, rattan and/or the like.

In the continuous reactor, some of the yeast is immobilised on the carrier and some of it may be freely suspended. Conventional known brewing yeasts are well suited for use in such a reactor. However, if highly flocculable yeasts are used, a high yeast concentration will be quickly reached in the reactor, and the high yeast concentration is also maintained, thus improving the efficiency of the reactor.

The immobilisation of yeast can be implemented in any known way, e.g. as described in patent specification US 4915959.

The amount of immobilised yeast in the reactor may vary as is known in the art, a preferable amount being 10^6 - 10^9 yeast cells/ 1 cm^3 of filler particles. The service life of the wooden particles used for yeast immobilisation is a few months, e.g. 1 - 6 months, but it may be as long as 1 year or more.

The rate at which the unmaturred beer flows through the reactor and its retention time in the reactor have an effect on the diacetyl content of the beer. The flow rate of the unmaturred beer is adjusted to a value such that a sufficient amount of diacetyl is reduced to acetoin in the reactor, with the result that the diacetyl concentration in the matured beer does not exceed an acceptable taste threshold. The flow rate of unmaturred beer through the reactor may be 0.05 - 2 times the reactor volume / h. A preferred flow rate of unmaturred beer is of the order of 0.5 - 1 reactor volume / h. The temperature in the reactor is 5 - 22 °C, preferably 5 - 20 °C. Even higher temperatures may be used.

The maturation reactor may be pressurised to maintain the carbon dioxide in a dissolved state in the reactor. Free carbon dioxide may hamper the operation of the reactor. The operation pressure can be se-

00423004 043400

WO 98/49264

PCT/FI98/00370

7

lected according to temperature, desired taste and beer quality.

After the maturation, the beer can be cooled to a desired stabilising temperature, and secondary
5 treatment of the beer, such as stabilising, filtering and decanting, can be implemented in a manner known in itself.

Because of their low price, the wooden particles and/or similar particles used as filler may be
10 thrown away after use. Disposal of the particles is easy and free of risks. The filler may also be regenerated after use, e.g. by treating them with hot water or vapour, by washing or by some other suitable treatment.

15 If desirable, the wooden particles and/or similar particles used as filler can be subjected to a treatment prior to immobilisation. The particles can be e.g. washed or treated in some other way as desired.

20 The continuous maturation reactor of the invention is an upright column in which the liquid flows through the column from bottom to top or from top to bottom. The diameter of the reactor is of the order of $1.5 \pm 1 - 2.5 \pm 1$ m and its height is of the order of
25 $2.5 - 10$ m. The column may be provided with one or more sieves, intermediate bottoms or flanges to keep the filler particles in the reactor. The column is filled mainly with wooden particles and/or similar particles with yeast immobilised on them.

30 As compared with prior art, the advantages of the invention are based on the use of a cheaper carrier material, which gives the same final result as more expensive carrier materials.

35 The low price of the wooden particles and/or similar particles also makes it unnecessary to regenerate the particles. When expensive carriers are used, regeneration is necessary to prolong the service life

004400404400

WO 98/49264

PCT/FI98/00370

8

of the carrier. Regeneration causes direct and indirect additional costs.

Wood and/or similar material also has the advantage that, being a natural material, it is free of risks.

The invention will now be described in detail via the following examples.

EXAMPLE 1

10

Test arrangements:

Räuchergold KL1 beech chips (5 litres) were cooked in ion-exchanged water (5.5 litres) for an hour. The water was removed and the chips were cooked for 4 hours in ethanol containing 10 % alcohol by volume. The alcohol solution was removed and finally the chips were cooked for 1 hour in ion-exchanged water.

The reactor was filled with the wet chips up to the 5.1 l mark. The reactor was assembled and autoclaved at 121 °C for 21 minutes together with the connections and hoses. After cooling, 3 litres of yeast suspension was pumped into the reactor in 6 hours by using a hose pump. Air was supplied into the reactor at the rate of 50 ml/min and wort at the rate of 100 ml/h overnight at 20 °C. After this, the supply of materials was stopped and the reactor was cooled to 10 °C.

The unmatured beer fed into the process was unmatured beer produced via immobilised main fermentation, in which the total content of visinal diketones was about 0.8 - 0.3 mg/ml. After the main fermentation, the unmatured beer was filtered through Seitz K filter paper into an autoclaved (121 °C, 20 min) restaurant container, which was used as a supply container for the secondary fermentation reactor.

WO 98/49264

PCT/FI98/00370

9

Description of the process:

The process comprises heat treatment of unmatured beer, its cooling to 10 °C, secondary fermentation (maturation) with immobilised yeast, and reception of the product.

From the supply container, the unmatured beer is pumped into heat treatment using a diaphragm pump (Prominent Mini Gamma). The heat treatment (80 °C, about 60 min) takes place in a thin-walled metal retention pipe immersed in a water bath at about 80 °C. The beer removed from the heat treatment flows into a cooling jacket made of glass, where it is cooled to the secondary fermentation temperature of 10 °C. The cooled beer flows through the reactor from bottom to top. From the top of the reactor, the beer flows via a separating funnel into a receiving container. The receiving container used is a 50-l restaurant container.

Analyses:

From the unmatured beer fed in, from the heat treated unmatured beer and from the post-fermented beer, the total amounts of visinal diketones (total VDK), free diketones (free VDK), aromatic substances and apparent extract concentration were analysed. The retention time in the reactor was estimated based on the flow rate. In addition, the beer colour was analysed twice during the test period.

Results:

The retention times in the reactor are presented in Table 1. With the reactor filled up to the 5.1 l mark, the liquid volume in the reactor was 3.6 litres. The internal amount of liquid within the chips, which is very small as the chips are wet all the time, was not taken into account, nor was the liquid remaining on the surface of the chips.

WO 98/49264

PCT/FI98/00370

10

chips, which is very small as the chips are wet all the time, was not taken into account, nor was the liquid remaining on the surface of the chips.

5

Table 1.

Flow rate	Retention time per volume of carrier material	Retention time according to the amount of liquid	Heat treatment time
ml/h	h/carrier volume	h	min
200	25.5	18.0	65
300	17.0	12.0	43
400	12.8	9.0	32

Tables 2 - 4 present the conversions of visinal diketones for different flow rates.

10

Table 2. Concentrations of visinal diketones (mg/dm³) and their conversion (%) at flow rate 200 ml/h.

1. determination	Supply	Heat treated	Post- fermented	Conver- sion
total diacetyl	0.77	0.70	0.02	97.4
free diacetyl	0.54	0.75	0.02	96.3
total pentanedione	0.20	0.18	0.01	95.0
free pentanedione	0.14	0.17	0.00	100.0
total VISINAL DIKETONES	0.97	0.98	0.03	96.9
2. determination				
total diacetyl	0.41	0.39	0.02	95.1
free diacetyl	0.23	0.36	<0.01	
total pentanedione	0.13	0.11	<0.01	
free pentanedione	0.08	0.10	<0.01	
total VISINAL DIKETONES	0.54	0.50	<0.03	

00423004 012400

WO 98/49264

PCT/FI98/00370

11

free pentanedione	0.07	0.10	<0.01	
total VISINAL DIKETONES	0.32	0.33	<0.03	90.6

Table 3. Concentrations of visinal diketones (mg/dm³) and their conversion (%) at flow rate 300 ml/h.

5

1. determination	Supply	Heat treated	Post-fermented	Conversion
total diacetyl	0.28	0.27	0.01	96.4
free diacetyl	0.17	0.27	0.01	94.1
total pentanedione	0.14	0.13	0.01	92.9
free pentanedione	0.07	0.12	<0.01	
total VISINAL DIKETONES	0.42	0.40	0.02	95.2
2. determination				
total diacetyl	0.39	0.37	0.02	94.9
free diacetyl	0.23	0.39	0.02	91.3
total pentanedione	0.22	0.19	0.01	95.4
free pentanedione	0.11	0.18	<0.01	
total VISINAL DIKETONES	0.61	0.56	0.03	95.1

Table 4. Concentrations of visinal diketones (mg/dm³) and their conversion (%) at flow rate 400 ml/h.

	Supply	Heat treated	Post-fermented	Conversion
total diacetyl	0.46	0.41	0.07	84.8
free diacetyl	0.27	0.38	0.06	77.8
total pentanedione	0.19	0.16	0.01	94.7
free pentanedione	0.09	0.14	0.01	88.9
total VISINAL DIKETONES	0.65	0.57	0.08	87.7

0042004 4002460

WO 98/49264

PCT/FI98/00370

12

Table 5 presents the average changes in the aromatic substances in the process as a percentage of the initial value. Table 5 shows that only the acetaldehyde concentration has changed significantly during the process. This is in fact a favourable change because an excessive acetaldehyde content would give the beer a solvent-like flavour. The results are the average values for three determinations at different flow rates.

10

Table 5.

	Supply	Heat treated	Post-fermented
Aromatic substance	%	%	%
ethyl acetate	100	97	99
3-methyl butyl acetate	100	69	80
propanol	100	101	102
2-methyl propanol	100	100	102
3-methyl propanol	100	99	101
2-methyl butanol	100	99	101
acetaldehyde	100	103	68

Table 6 presents the results of the determinations of apparent extract concentration and colour. The apparent extract concentration and colour of the beer were determined twice during the test period to make sure that no changes occurred in the fermentation and that the darkish wood imparted no colour to the beer.

20

Table 6.

	Supply	Heat treated	Post-fermented
extract concentration 200 ml/h (%)	2.28	2.26	2.22
extract concentration 300 ml/h (%)	1.91	1.98	1.98

00423004 043400

WO 98/49264

PCT/FI98/00370

14

CLAIMS

1. Continuous method for the maturation of beer after main fermentation, in which method the un-matured beer, after removal of yeast and a heat treatment, is passed into a bio-reactor filled with a carrier material with yeast immobilised on it, characterised in that the carrier material mainly consists of wooden particles and/or similar particles.

2. Method as defined in claim 1, characterised in that said particles are chip-like or stick-like particles or particles shaped like any regular or irregular bodies, whose dimension is of the order of 1 - 100 mm, advantageously 1 - 50 mm, preferably 2 - 20 mm.

3. Method as defined in any one of claims 1 - 2, characterised in that the wooden particles have been produced from deciduous wood.

4. Method as defined in any one of claims 1 - 2, characterised in that the wooden particles have been produced from coniferous wood.

5. Method as defined in any one of claims 1 - 2, characterised in that the wooden particles have been produced from tropical gramineous plants.

6. Method as defined in any one of claims 1 - 5, characterised in that the yeast used in the reactor is conventional brewing yeast and/or highly flocculable yeast.

7. Method as defined in any one of claims 1 - 6, characterised in that the amount of yeast in the reactor is 10^6 - 10^9 cells/l cm^3 of particles.

8. Method as defined in any one of claims 1 - 7, characterised in that the temperature in the reactor is 5 - 25 °C, preferably 5 - 20 °C.

9. Method as defined in any one of claims 1 - 8, characterised in that the flow rate of

unmatured beer through the reactor is of the order of 0.05 - 2 times the reactor volume / h, preferably 0.5 - 1 reactor volume / h.

10. Method as defined in any one of claims 1 - 9, characterised in that the particles are regenerated, preferably using hot water or steam.

11. Method as defined in any one of claims 1 - 10, characterised in that the particles are subjected to a treatment, preferably a water cooking treatment or ethanol extraction treatment, prior to immobilisation.

12. Method as defined in claim 11, characterised in that the particles are washed.

13. Continuous beer maturation reactor, which is an upright column-type flow-through reactor containing one or more sieves, intermediate bottoms or flanges and which is filled with a carrier material with yeast immobilised on it, characterised in that the carrier material mainly consists of wooden particles and/or similar particles.

14. Maturation reactor as defined in claim 13, characterised in that said particles are chip-like or stick-like particles or particles shaped like any regular or irregular bodies, whose dimension is mainly of the order of 1 - 100 mm, preferably 1 - 50 mm.

09123004 042400

COMBINED DECLARATION FOR PATENT APPLICATION AND POWER OF ATTORNEY

As a below named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated below next to my name.

I believe that I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled _____
Method for the maturation of beer

the specification of which: (check one)

- ☐ is attached hereto;
☒ was filed as U.S. application Serial No. 09/423,004
on 29 Oct.1999 and was amended by Preliminary Amendment
on _____ (if applicable);
☐ was filed as PCT international application No.
_____ on _____ and was amended under
PCT Article 19 on _____ (if applicable).

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment(s) referred to above.

I acknowledge the duty to disclose to the USPTO all information known to me to be material to patentability as defined in Title 37 Code of Federal Regulation §1.56(a).

I hereby claim foreign priority benefits under Title 35 United States Code §119 of any foreign application(s) for patent or inventor's certificate or of any PCT international application(s) designating at least one country other than the United States of America listed below and have also identified below any foreign application(s) for patent or inventor's certificate or any PCT international application(s) designating at least one country other than the United States of America filed by me on the same subject matter having a filing date before that of the application(s) of which priority is claimed:

PRIOR FOREIGN/PCT APPLICATION(S)
AND ANY PRIORITY CLAIMS UNDER 35 U.S.C. 119:

PRIORITY
CLAIMED

Finland	971838	29/04/1997
(Country)	(Number)	(Day/Month/Year Filed)
(if PCT indicate PCT)		

[x]	[]
Yes	No

(Country)	(Number)	(Day/Month/Year Filed)	[] Yes	[] No
(if PCT indicate PCT)				

(Country)	(Number)	(Day/Month/Year Filed)	[] Yes	[] No
(if PCT indicate PCT)				

I hereby claim the benefit under Title 35 United States Code §120 of any United States application(s) or PCT international application(s) designating the United States of America that is/are listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in that/those prior application(s) in the manner provided by the first paragraph of Title 35 United States Code §112, I acknowledge the duty to disclose material information as defined in Title 37 Code of Federal Regulations §1.56(a) which occurred between the filing date of the prior application(s) and the national or PCT international filing date of this application:

PRIOR US/PCT APPLICATION(S)
FOR BENEFIT UNDER 35 U.S.C. 120

PCT/FI98/00370	28/04/1998	
(Application No.)	(Filing Date)	(Status - Patented, Pending, or Abandoned)
<hr/>		
(Application No.)	(Filing Date)	(Status - Patented, Pending, or Abandoned)
<hr/>		
(Application No.)	(Filing Date)	(Status - Patented, Pending, or Abandoned)

As named inventor, I hereby appoint the following attorney(s) and/or agents, to prosecute this application and transact all business in the Patent and Trademark Office connected therewith:

Glenn O. Starke	<u>17,031</u>	Andrew S. McConnell	<u>32,272</u>
Eugene R. Sawall	<u>17,431</u>	Joseph J. Jochman, Jr.	<u>25,058</u>
Daniel D. Fetterley	<u>20,323</u>	Casimir F. Laska	30,862
George H. Solveson	<u>25,927</u>	Peter C. Stomma	36,020
Gary A. Essmann	<u>29,376</u>	Edward R. Williams, Jr.	<u>36,057</u>
Thomas M Wozny	<u>28,922</u>	William L. Falk	<u>27,709</u>
Michael E. Taken	<u>28,120</u>		

Address all Telephone Calls to:

Daniel D. Fetterley
(414) 271-7590

Address all correspondence to:

ANDRUS, SCEALES, STARKE & SAWALL
100 East Wisconsin Ave., Ste. 1100
Milwaukee, Wisconsin 53202
(414) 271-7590

1-00

[illegible]

[Handwritten signature]

40

-3-